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Research Article

In vivo Antifertility and Safety Profiles of Kenyan *Moringa oleifera* Lam. (Moringaceae) extracts

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Background: Unsustainable high population growth rate coupled with many women dying of complications of unsafe abortion, due to a large number of unwanted pregnancies, has been a challenge in many parts of the world especially in developing countries. This indicates that new and alternative contraceptive methods that are safe, cheap and convenient are needed. *Moringa oleifera* Lam (Moringaceae) was selected for this study based on previous studies that indicated antifertility effect in rats, of the aqueous extract of the roots and the stem bark.

Objective: To establish the antifertility properties of *M. oleifera*.

Methodology: The aerial parts, seeds, root bark and twigs were extracted using methanol, petroleum ether, dichloromethane and ethyl acetate. *In vivo* antifertility evaluations in Swiss female mice, acute and sub-chronic toxicity and phytochemical studies were carried out on *M. oleifera* extracts.

Results: The ethyl acetate extract of the seeds of *M. oleifera* demonstrated reversible antifertility effect at 800mg/kg. Physiological tests carried out on mice revealed that the extract arrested the estrus cycle either at the diestrus or the proestrus phase by prolonging them. Acute and chronic toxicity evaluation of the extract at 800mg/kg established the safety at the tested concentration. Thin layer chromatography (TLC) of the extract revealed the presence of terpenoids, steroids and fluorescent compounds, which may be responsible for the antifertility effect that was observed.

Conclusion: The findings validate the ethnomedicinal use of *M. oleifera* seeds through the establishment of its safety, and the antifertility properties that make the extract a potential source of an alternative herbal contraceptive through further studies and development.

Key words: *Moringa oleifera*, antifertility effect, estrus cycle, toxicity, phytochemical profile.

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1. Introduction

Each year, over half a million women die of complications of pregnancy, childbearing, and unsafe abortion. A vast majority of the deaths occur in developing countries (Ziraba et al, 2009; WHO, 2014; MoH, 2013). It is estimated that between 10% and 20% of these pregnancies are unwanted at the time of conception. Thus, up to 100,000 maternal deaths could be avoided if women who did not want children used effective and acceptable contraceptives (Datey et al, 1995; WHO, 1997; WHO, 2003; Westhoff et al, 2007; WHO, 2012[a]; 2012[b]). Lack of use, improper use or discontinuation of family planning methods has led to a high population growth rate in many countries hence causing an imbalance between populations and the available resources (Umadevi et al, 2013). In Kenya, abandonment of contraceptive use while still in need of contraception and contraceptive failure account for about 65% of all discontinuations (APHRC, 2001; GoK,

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1991; WHO, 2012[b]). Consequently, Kenya is currently grappling with a huge population outburst of about 40 million people (MSPND, 2010). This situation is in dire need of an intervention in order to reduce the number of deaths being caused by abortions performed by unqualified personnel as well as to reduce the population. Traditional birth control method by the use of herbal remedies has been in existence for many years among various societies (Jain et al, 2004; Kokwaro, 2009; Umadevi et al, 2013). Consequently, various studies have established different antifertility activities of medicinal plants that include anti-implantation, abortification, ecobolic, oestrogenic and spermicidal effects (Priya et al, 2012; Kassem et al, 2005; Tafesse et al, 2005; Ganguly et al, 2007; Keshri et al, 2007). This study therefore aimed at investigating the antifertility properties of *Moringa oleifera*. Lam (Moringaceae). Studies on *M. oleifera* growing in other parts of the world have shown that aqueous extract of the roots and the stem bark, had antifertility effect in rats. It exhibited estrogenic, antiestrogenic, progestational and antiprogestational activities (Shukla et al, 1988; Zade & Dabhadkar, 2015). The antifertility effect of Kenyan *M. oleifera* extracts is largely unknown. *M. oleifera* is a widely distributed plant that can survive humid tropics and hot, dry land. In Kenya, it is found in Nyanza, Eastern and Coastal regions (Beentje et al, 1994). The tree is highly nutritious and has been reported to have several ethnomedical uses including birth control (Fahey, 2005; Anwar et al, 2007).

2. Methodology

2.1 Collection of plant materials

The plant materials were collected from Kibwezi in Makueni County and identified by Mr. Mathias Mbale, a plant taxonomist at the East Africa Herbarium-National Museums of Kenya, Nairobi, where the voucher specimen was deposited (*M. oleifera* voucher number **EK710**). The plant material [aerial parts (leaves & flowers), seeds, root bark and twigs] were separately dried under room temperature for 2 weeks then pulverized using an electrical mill. The weight of the various parts was taken after drying.

2.2 Extraction

2.2.1 Organic extraction

The ground plant material (50-100 g) from each plant part were extracted twice with methanol, petroleum ether, dichloromethane and ethyl acetate (approx. 200 ml) by soaking them for 48 h in the respective solvent. Samples were then filtered using Whatman® (No.1) filter paper. The filtrates were then dried under reduced pressure using a rotatory evaporator (Buchi R114) at controlled temperature of between 40-60^{o}C and then transferred into weighed vials (Mallikharjuna et al, 2007). The percentage yield was then calculated and recorded.

2.2.2 Aqueous extraction

Fifty grams of the ground plant material were weighed and transferred into a conical flask. The materials were then covered with distilled water (100 ml) and placed in the water bath at 60^{o}C for 2 h. The samples were then filtered and freeze-dried using a freeze drying machine (Modulyo EF4). The percentage yield was calculated and recorded.

2.3 *In vivo* bioassays

2.3.1 Animal handling

The research was conducted in accordance with the internationally accepted principles for laboratory animal use and care as found in the NIH care and use of animals' guidelines. Ethical clearance was sought from KEMRI's Scientific Steering and the Ethical Review Committees. A total of 300 (including repeat experiments) healthy Swiss mice of both sexes, 8-10 weeks old weighing 18-22 g, randomly bred at the KEMRI animal house were used in the study. The animals were housed spaciously in standard polypropylene cages clearly labeled with experimental details. The mice were maintained at room temperature and 60-70% relative humidity range appropriate for their species, with 12 h cycle of day and night light conditions. This enabled them to acclimatize with minimal stress and physiological alteration. The mice were fed on commercial rodent food and water *ad libitum.* A canular was used for oral drug administration. The size of the needle that was used to draw blood was 25 gauges. After the experiment, the mice were euthanized. The euthanized mice were then placed in biohazard disposable bags for incineration.

2.3.2 Drug constitution

On the day of extract administration, each of the organic extract was freshly prepared by dissolving in a solution consisting of 70% Tween 80, 30% ethanol and diluted with double distilled water. The aqueous extracts were prepared using distilled water (Houghton & Raman, 1998). A conventional drug (Femiplan®, levonogestrel: 0.15 mg & ethinylestradiol: 0.03 mg) was used as the positive control. The effective concentration of the femiplan was determined by administering three doses of 180, 90 and 45 mg/kg to determine the lowest effective dose of the drug in mice. Both 180 and 90 mg/kg were effective in controlling the fertility of the mice. However, 45 mg/kg concentration was not effective in controlling the fertility of the mice. In the subsequent experiments, 90 mg/kg was taken to be the lowest effective dose and was used as the positive control.

2.3.3 Fertility test

Fifty female mice were divided into 5 mice per cage. In each cage a male was introduced to the female mice and were allowed to mate. After two weeks, the male mice were withdrawn after pregnancy sign (bulging stomach) was noted in some of the female mice. The expectant females delivered and the number of pups noted. Those that did not conceive were separated from the pregnant mice and were considered infertile at the time and were not used in the subsequent experiments (Mali et al, 2002).

Screening for the anti-fertility effect in mice

This study was carried out according to the method described by Ganguly et al (2007). Eleven groups (each group containing 3 fertile female mice) were selected for the study. Eight groups received different extracts while 3 groups received distilled water, Tween 80 and

femiplan as controls. The drug administration was first carried out for 8 days before the introduction of males. All the experimental mice were then allowed to mate with the identified fertile male mice, and the drug administration continued for 21 days. The number of litters was determined after the completion of one gestation period of 21 days in all experimental groups. The study was independently repeated and the average of the two studies determined.

Reversibility test

The reversibility of the anti-fertility effect of the extracts was also studied in the treated groups according to the method described by Salhab et al (1997). Briefly, the active extract was administered at 800 mg/kg continuously to a group of 3 mice for 21 days, and then withdrawn. After 21 days of extract withdrawal, animals were allowed to mate with male mice. The number of litters was determined after the completion of one gestation period. The study was independently repeated and the average of the two studies determined.

Effect of the ethyl acetate extract of the seeds of *M. oleifera* on the estrus cycle

Five fertile female mice were employed for the study. Vaginal smears from each animal were examined under a microscope once per day every morning between 9.00 and 10.00 am for 21 days. This accounted for about 4 - 5 cycles. Each mouse was held in a supine position and the vaginal secretion was collected after cleansing with 0.2 ml of normal saline (NaCl 0.9%) contained in a smooth plastic pipette and placed in a tube (Marcondes et al, 2002). A small drop of the cell suspension was then placed on a glass slide and examined under a light microscope at x10 and x40 magnification. The cell suspension was evaluated to determine the phases of estrus cycle using the proportion of characteristic cell types such as the leucocytes, cornified and epithelial cells (Abu & Uchendu, 2011; Malaivijitnond et al, 2006). The duration of the estrus cycle together with that of the various phases was determined as described by Makonnen et al (1997) for 21 days. Administration of the test drug (ethyl acetate extracts of the seeds at 800 mg/kg) followed and the same parameters were determined.

Effect of the ethyl acetate extract of the seeds of *M. oleifera* on the weight of genital organ and body weight

The experiment was done according to Makonnen et al (1997) with some few modifications. Five groups of 5 female mice in each group were employed. The experimental mice received the test extract at 800 mg/kg and the control groups received distilled water and 10% Tween 80, for 10 days through the oral route. On the 11th day, all the animals in all the groups were weighed and sacrificed. The ovaries and uteri were resected, separated from the surrounding tissues, and then blotted on aluminum foils (Gebrie et al, 2005). The organs were weighted and a ratio calculated by dividing the weight of the ovary, as well as, that of the uterine in milligrams by body weight in grams. The rise in the uterine ratio was an indication of the estrogenic effect of the extract as described by Vogel (1997).

2.3.4 Acute toxicity assay

The acute toxicity effect of the bioactive extracts was carried out according to the method described by Mukinda and Syce (2007). Briefly, 8 groups consisting of 5 female mice per group were used in this experiment. The doses of the bioactive extracts administered in single doses orally were 0, 1000, 2000, 3000, 4000 and 5000 mg/kg. The general behavior of the mice was continuously monitored for 1 h after dosing, periodically during the first 24 h especially the first 4 h (Hilaly et al, 2004), and then daily thereafter, for a total of 14 days. Changes in the normal activity of mice and their weights were monitored and the time at which signs of toxicity or death appeared recorded.

2.3.5 Sub-chronic toxicity study in mice

The study was carried out according to Mukinda and Syce (2007) with some few modifications. Female mice were housed in 5 (I-V) groups of 5 animals. The mice were first left for 7 days to acclimatize to laboratory conditions. The extracts were administered orally, daily for 3 months, to the groups I to VI at treatment doses of 0, 100, 1000 and 2000 mg/kg respectively. In the treated animals, differences in their normal behaviour, fur condition, discharge, movements and mortality of the mice were monitored daily. Changes in body weight and food intake were recorded weekly. At the end of the 3 month-experiment, all the animals were euthanized and blood samples collected through cardiac puncture into EDTA containing tubes and in plain tubes for haematological and biochemical analyses respectively. The collected blood samples were analyzed using a hemoanalyzer machine (URIT-3300).

2.4 Phytochemical screening of the bioactive extract

Phytochemical screening of the extracts of the seeds of *M. oleifera* was carried out using thin layer chromatography (TLC) to determine the presence of secondary metabolites such as steroids, terpenoids, tannins, flavonoids, alkaloids, saponins, glycosides and reducing sugars (Harborne, 1998; Mallikharjuna et al, 2007).

2.5 Statistical analysis

The litter number and weight of the uterus and body weight ratio were expressed as mean ± standard deviation (S.D). For the hematological and biochemical parameters, a significant difference between control and experimental groups was assessed by the use of Student's t-test while the analysis of variance (ANOVA) was used to assess any significant difference among the groups. The level of significance was set at ρ values less than 0.05.

3. Results and Discussion

Collection and extraction

The weights of the various plant parts of *M. oleifera* (voucher number **EK710**) were: aerial parts (leaves & flowers) - 1.45 Kg, seeds – 1 Kg, root bark – 0.62 Kg and twigs – 0.31 Kg. Extraction was carried out using dichloromethane, methanol, ethyl acetate and water. The

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percentage yields of the extracts are summarized in **Table 1**. Extraction of the root bark and seeds using dichloromethane resulted in the highest yields of extracts (**Table 1**).

Table 1: Percentage yields (%) of the *M. oleifera* extracts

Part	PE	CH_2Cl_2	AcOEt	MeOH	H_2O
Aerial parts	1.4	nd	1.04	3.32	nd
Root bark	0.73	1.08	0.36	3.43	13.36
Seeds	13.68	19.87	15.01	1.56	11.08
Twigs	0.4	1.78	1.06	4.80	

PE - petroleum ether; CH_2Cl_2 – dichloromethane; MeOH – methanol; H_2O – water; nd – not done

Fertility Test

Determination of the fertility in the female mice gave a rate of 60%. The results of fertility test suggested that some of the mice were not receptive for the 21 days that the male mice were in the cage, and thus did not give birth during the first gestation period. The litter size per mouse recorded an average of 6. The mice that gave birth were considered as fertile and were used in the screening for the antifertility activity of the candidate plants.

Antifertility and reversibility effect

Ethyl acetate extract of the seeds of *M. oleifera* demonstrated the highest antifertility effect (**Table 2**) since the female mice did not bear offspring after exposure to the male.

On withdrawal of the extract, the female mice on exposure to the male, conceived and bore an average of 6 pups, a value comparable to the positive control (mice administered with femiplan) and the negative control (fertile mice not treated, **Table 3**), indicating that the antifertility effect of the extract is reversible. In previous studies antifertility effect was reported in the aqueous and alcohol extracts of the roots and the stem bark of *M. oleifera* respectively (Shukla et al, 1988; Prakash et al, 1987; Zade & Dabhadkar, 2015). However, no reports have indicated that the seeds have an antifertility effect. Ethanol and aqueous extracts of the seeds have been found to have other biological activities such as antimicrobial, antitumor, antioxidant activities amongst others (Guevara et al, 1999; Fahey, 2005; Anwar et al, 2007).

Table 2: Fertility of female mice after 21 days of treatment with 800mg/kg of test extracts (Male: Female ratio, 1:3)

Extract	Solvent	Extract dose mg/kg	No. of fertile/treated mice	Litter size ±S.D
M. oleifera (seed)	Pet ether	800	2/3	8.00±1.0
	Ethyl acetate	800	0/3	0
	DCM	800	3/3	9.00±0.58
	Water	800	3/3	6.67±0.33
M. oleifera (Twigs)	Methanol	800	3/3	7.33±0.33
M. oleifera (Root bark)	Methanol	800	3/3	6.00±0.58
	Water	800	2/3	6.00±0.50
M. oleifera (aerial)	Methanol	800	3/3	6.00±0.58
Negative control	Water	N/A	3/3	8.00±0.58
	Tween 80	10%	2/3	6.00±1.00
Positive control (Femiplan™)		90	0/3	0

The data is an average of two independent experiments.

Table 3. Fertility of female mice after 21 days of treatment after withdrawal of the treatment during a repeat of the experiment

Plant & +ve control	Solvent	Extract/Drug Dose mg/kg	No. of fertile/ treated mice	Total no. of pups per group	Litter size ± S.D
M. oleifera (seed)	Ethyl Acetate	800	3/3	16	5.3±1.41
Femiplan	Water	180	3/3	20	6.7±0.58
Femiplan	Water	90	3/3	22	7.3±0.58

The data is an average of two independent experiments.

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Effect of the ethyl acetate extract of the seeds of *M. oleifera* on the estrus cycle

The estrus cycle in mice occurs over 4 - 5 days in 4 stages: the proestrus stage (pre-ovulatory day) and the ovulatory stages, which include estrus, metestrus and diestrus stages defined by different forms of epithelial, cornified and leukocyte cells (Caligioni, 2009). Observation of the vaginal smears using microscopy provides information on the stage and the length of the cycle of a mouse (Makonnen et al, 1997). During treatment of the female mice with the ethyl acetate extract of the seeds, the length of the estrus cycle in all the treatment groups was significantly changed as compared with the control group that received distilled water. Mice in the control group exhibited a regular estrus cycle of 4 to 5 days. The extract arrested the normal estrus cycle at either the diestrus or the proestrus phase. The diestrus and proestrus phases of the cycle of the treated mice were prolonged in the treated groups. However, the estrus and metestrus phases of the treated mice were highly reduced in the treatment group. Similar observations of prolonged estrus and diestrus phases (by 48 h) were observed when albino rats were administered with the alcohol extract of the stembark of *M. oleifera* in a study carried out by Zade & Dabhadkar, 2015. During the administration of ethyl acetate extract of *M. oleifera* seeds, the mice exhibited diestrus phase for most of the days, followed by proestrus phase and only one day for some of the mice exhibited the estrus and metestrus phases. The disruption of the estrus cycle is a likely mechanism of action of the *M. oleifera* seed extract in mice and should further be investigated.

Effect of the extract on the weight of genital organ and body weight

Generally, the ethyl acetate extract of the seeds of *M. oleifera* did not have a suppressive effect on the ovary as well as the uterus (**Figure 1**). At a concentration of 800mg/kg the extract administered for 10 days did not affect the uterine fresh weight significantly (ρ = 0.077), as well as, the fresh weight of the ovaries (ρ = 0.145). This indicates that even though the seed extract inhibits conception in mice, the mechanism of action of the extract does not involve altering of the physiology of the ovaries or the uterus.

Acute toxicity assay

Interestingly, administration of the ethyl acetate extracts of *M. oleifera* seed showed no death or signs of toxicity up to 4000mg/kg. However, at the highest tested dose (5000mg/kg) there were mild signs of toxicity that included hypo-activity, low appetite, and piloerection. This observation was comparable to the negative control. It is worth noting that at 5000mg/kg the toxicity was noted for 1 h however, after the 2nd h after extract administration, most of the mice had regained their normal activity, as well as appetite. Keeping in mind that the antifertility active concentration of the extract is 800mg/kg, it can be deduced that the plant extract is safe at the active concentration. Previous reports indicated that the alcoholic extract of the stem bark of *M. oleifera* did not exhibit acute toxicity in rats (Zade & Dabhadkar, 2015).

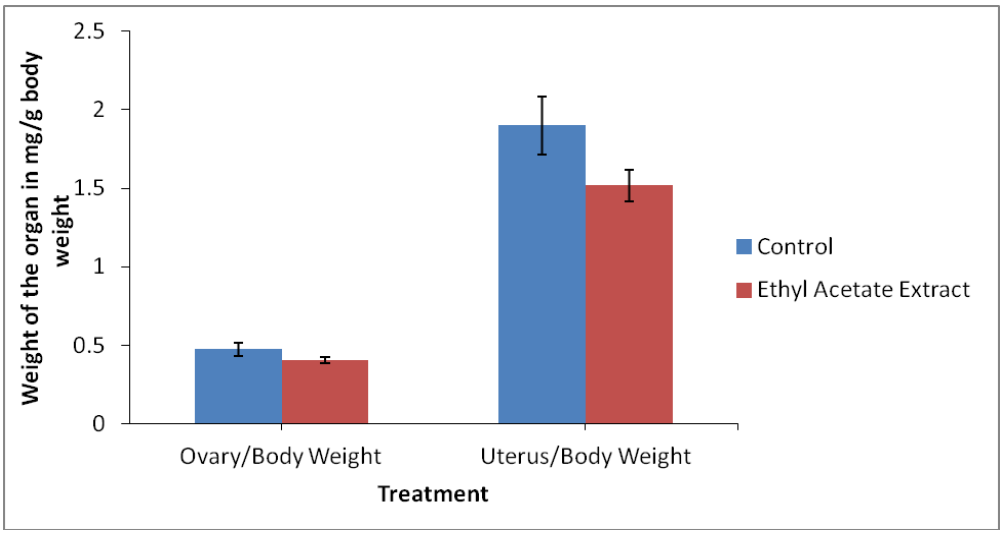

Figure 1: The effect of the ethyl acetate extract of the *M. oleifera,* (seed) in comparison with the control on wet weight of mice uterus and the ovaries

Sub-chronic toxicity study in mice: Effect of oral *Moringa oleifera* ethyl acetate extract of the seeds, on the general behavior of mice

Oral administration of the ethyl acetate extract of the seeds of *M. oleifera* caused no noticeable change in the general behavior of the mice for the first two months of extract administration. However, the fur, movement and the general activity of the mice differed from the control group. Interestingly, the weight of the mice that received the extract at 100mg/kg, was higher than that of the control group (**Figure 2**). This may have been observed due to the nutritional benefits associated with the *M. oleifera* seeds at the given concentration (Fahey, 2005; Anwar et al, 2007). The amount of food intake between the two groups (control and those administered with 100mg/kg of extract) was comparable. On the other hand, the mean weight of the mice that received 1000mg/kg and 2000mg/kg extract dropped, compared to the control group, an indication that administration of a high dose of the extract may have affected the appetite of the mice, hence causing a reduction in food-intake. Similar observations were made in a study by Adedapo et al 2009, where a drop in the weight of rats was reported after administering an aqueous extract of *M. oleifera* leaves; the weight reduction increased with an

increase of the dose administered (400 - 1600mg/kg). In addition to a weight reduction in this study, the mice manifested minor symptoms of toxicity comprising rough fur and partial hypo-activity. However, no deaths occurred at any of the doses (up to 2000mg/kg) administered indicating that the LD_{50} for chronic oral dosing with *M. oleifera* extract was much higher than 2000mg/kg. This further confirms that the plant extract is safe at the active concentration (800mg/kg), however, further histopathological investigations need to be carried out to conclusively confirm that the extract is safe in mice at the given concentration.

Sub-chronic toxicity study in mice: **Effect of oral *Moringa oleifera* ethyl acetate extract on the hematological and biochemical parameters of mice**

The parameters analyzed included white blood cells (WBC), red blood cells (RBC), hemoglobin, hematocrit and platelet levels. All the parameters for the treated mice and the negative control were comparable. For instance, the WBC levels in groups that received 100mg/kg, 1000mg/kg and 2000mg/kg were 7.280 x 10^9, 6.920 x 10^9 and 7.600 x 10^9/L respectively (**Table 4**). In the control group, the level of the WBC was 7.980 x 10^9/L. At ρ<0.05, significant difference in each group was checked versus the controls. There were no parameters in any test group that showed significant difference against the control hence the ethyl acetate extracts of *M. oleifera* seed can be said to be safe at 800mg/kg.

The hematological parameters, such as hematocrit, hemoglobin concentration, platelets, red and white blood cells in the treated mice did not differ significantly from those of the control group (**Table 4**). This is in agreement with published hematological parameters that were determined for adult Balb/c mice, but with a few differences in the levels of WBC and platelets that may have occurred due to the genetic differences in the two species of mice (Nemzek et al, 2001). Hematological parameters are some of the targets for toxic compounds and may thus act as indicators of toxicity (Diallo et al, 2010). Thus the extract did not have effects on the circulating cells neither did it affect their production.

A significant difference of treatment-related changes was not observed in the levels of plasma analytes as compared to the control group (**Table 4**). However, a significant reduction in the levels of alanine transaminase (p = 0.0018 & 0.00055 at 100 & 2000mg/kg respectively), aspartate aminotransferase (p = 0.000016 at 2000 mg/kg) and total cholesterol (p = 0.0204 & 0.00095 at 1000 & 2000 mg/kg respectively) was noted. Elevated alanine transaminase and aspartate aminotransferase are biomarkers that predict toxicity and damage in the liver (Campion et al, 2013) however, lower values have not been reported to be a cause of liver toxicity but may serve as an independent predictive marker for increased long term mortality (Ramaty et al, 2014). The reduction of cholesterol at higher extract concentrations may have resulted from the changes in normal behavior in mice characterized by low appetite and hypo-activity rather than liver toxicity. It may also have been due to antiatherogenic effects resulting from metabolic effects that enhance glucose uptake as reported in other studies in which hypoglycemia effect of methanol and aqueous extracts of the leaves of *M. oleifera* was observed (Abd et al, 2014; Olayaki et al, 2015). It is worth noting that no significant difference was observed in creatinine level, which is an indicator of the kidney function (Campion et al, 2013). Based on the results presented here, it can be concluded that the ethyl acetate seed extract is safe at the effective dose.

Phytochemical screening

Thin layer chromatography (TLC) of extracts of the seeds of *M. oleifera* revealed the presence of various compounds or secondary metabolites. The test indicated the presence of terpenoids, alkaloids, phenols, glycosides and steroids in the various seed extracts. The ethyl acetate seed extract mainly consisted of terpenoids, steroids and fluorescent compounds. Compounds that have previously been isolated from the seeds of *M. oleifera* include: *O*-ethyl-4-(α-L-rhamnosyloxy)benzyl carbamate, 4(α-L-rhamnosyloxy)-benzyl isothiocyanate, niazimicin, 3-*O*-(6'-*O*-oleoyl-*β*-D-glucopyranosyl)-*β*-sitosterol, *β*-sitosterol-3-*O*-*β*-D-glucopyranoside, niazirin, *β*-sitosterol and glycerol-1-(9-octadecanoate), most of which are associated with various bioactivities (Faizi et al, 1998; Guevara et al, 1999; Anwar et al, 2007; Galuppo et al, 2013; Hussain et al, 2014). Hence the compounds already identified in the seeds of *M. oleifera* and others that may be present but unknown, can be further investigated for antifertility effect.

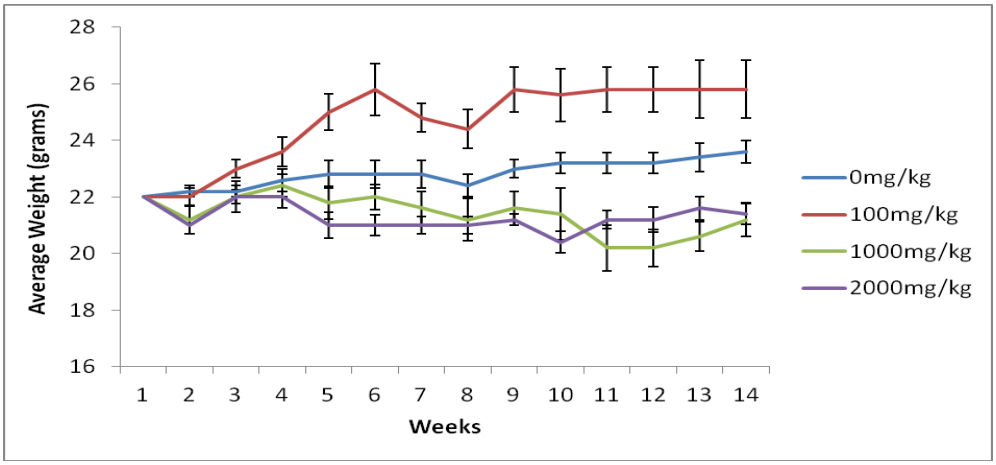

Figure 2: Changes in the body weight of mice after chronic oral treatment with an ethyl acetate extract of *M. oleifera* seed

Table 4: The effect of *M. oleifera* extract on the whole blood count and biochemical blood parameters of mice after 3 months daily oral dosing

Parameters	Dose of ethyl acetate extracts			
	0mg/kg	**100mg/kg**	**1000mg/kg**	**2000mg/kg**
White Blood Cells x 10^9/L	7.980±1.5803	7.280±0.7889	6.920±0.8737	7.600±0.4593
RBC x 10^{12}/L	8.9040±0.35004	9.0680±0.10131	8.7340±0.05464	8.7820±0.20984
Hemoglobin (g/dL)	14.700±0.4615	14.840±0.2694	14.360±0.1939	13.880±0.2596
Hematocrit (%)	46.660±2.1653	51.140±0.7194	49.120±0.7559	50.380±2.0781
Platelet x 10^9/L	443.20±55.356	430.00±31.420	467.00±9.721	493.40±44.002
Alanine transaminase IU/L	56±0.84	51±0.71*	57±0.71	49±0.95*
Aspartate aminotransferase IU/L	180±2.12	180±1.52	187±2.05	154±1.89*
Creatinine µmol/L	104±2.59	113±3.38	107±2.92	107±3.31
Total Cholesterol mmol/L	7.8±0.31	7.5±0.18	6.7±0.18*	5.5±0.13*

The data are expressed as mean ± S.E.M; AST activity levels increased with age (time); *values with $p < 0.05$

4. Conclusion

This study has established that the seeds of *M. oleifera* Lam. have antifertility properties and most importantly they are safe at the active concentration. The findings indicate that with further investigations the seeds have the potential to be developed as an affordable herbal contraceptive. It further validates the ethnomedical use of the plant amongst various indigenous communities.

Conflict of Interest

The authors declare no conflict of interest.

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